FINAL

Microbiological Sampling Report

for

National Oceanic & Atmospheric Administration

Samplings Conducted on the Sixth Floor
of Building SSMC-3
On February 17, 2000

Interagency Agreement #: D8H00CO31200
Task: 9903

May 16, 2000

Prepared by

US Public Health Service
Division of Federal Occupational Health
Bethesda Central Office

Executive Summary

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in rooms 6747 and 6855 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from these rooms and an indoor reference room 6745. Air samples were also collected from outdoors.

Findings are as follows:

· No fungal growth was detected from indoor Andersen samples. Indoor fungal spore levels, by Zefon sampler, were lower than those of outdoors.

· In general, fungal burden on surfaces was low.

· *Stachybotrys chartarum* was not detected from any air, wipe, or contact plate samples collected.

· Very low fungal burden was detected from wipe samples collected from surfaces of supply diffusers and return troughers in light fixture.

· Fungal levels in plenum, carpet, and furniture dust of these rooms were at 10^3 - 10^4 CFU/g of fine dust levels. *Stachybotrys chartarum* was detected from all samples except for the plenum dust collected from room 6747 and carpet dust from room 6855.

· A diverse fungal population was recovered from these dust samples. *Penicillium* and *Aspergillus niger* dominated dust samples collected from the ceiling plenum.

INTRODUCTION

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in rooms 6747 and 6855 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from these rooms and an indoor reference room 6745. Air samples were also collected from outdoors.

EVALUATION METHODOLOGY

Air Samples

Various types of samples were collected from these rooms on February 17, 2000. Two types of air samples were collected from each room: (1) culturable method using Andersenâ N-6 samplers at a flow rate of 28.3 L/min, and (2) non-culturable method using Zefonâ Air-O-Cell cassettes at a flow rate of 15 L/min. Indoor Andersenâ air samples were collected for 3 minutes and outdoor samples were collected for both one and three minutes. Two percent (2 %) malt extract agar (MEA) and cellulose Czapek agar (CCA) was used to recover general fungi and cellulose-loving fungi, respectively. Non-culturable air samples were collected at the aforementioned sampling locations. Indoor samples were collected for ten minutes and outdoor samples were collected for both five and ten minutes. Outdoor air samples were collected near the entrance of the building. Temperature and relative humidity measurements were collected from each air sampling location by a battery operated, direct readout Hygroskopâ meter.

Contact Plate Samples

To determine fungal burden on horizontal and vertical surfaces of these rooms, four to five contact plate samples were collected from each room. Samples were collected from randomly selected horizontal and vertical surfaces. Sampling was conducted by pressing the MEA-filled Rodacâ plate against the surface of interest for five seconds. A total of 14 contact plate samples were collected.

Swab Samples

Swab samples were collected from surfaces of each supply diffusers and return troughers in each room. They were collected by wiping a known area of surface with a sterile cotton swab (Culturette®) wetted with holding media. Approximately 5 in^2 area was wiped for return trougher and 4 in^2 for supply diffusers. The swab was then placed directly into its holder. Each holder was labeled with an identifiable number. A total of nine wipe samples were collected from these rooms.

Vacuum Dust Samples

Dust accumulated on carpeting, chairs and fabric system furniture, and the plenum were collected with a High Efficiency Particulate Air (HEPA) vacuum attached with a special "sock" device. For each carpet sample, a 3-ft by 3-ft area was vacuumed for at least five minutes. Total surface areas of 9 ft^2 were vacuumed from system furniture and chairs and composite as one sample. Dust accumulated above the ceiling plenum was also vacuumed and composite as one sample. One carpet sample, one composite furniture sample, and one composite plenum sample were collected from each room. A total of nine dust samples were collected.

All samples collected were sent for next morning delivery to FOH's Environmental Microbiology Laboratory (EML) in Philadelphia, Pennsylvania for analysis.

Laboratory Procedures

Upon receipt, all Andersen® air and contact plate samples were incubated in a 25°C incubator. Each swab sample was suspended in sterile distilled water, diluted serially, and inoculated onto agar plates. Both MEA and CCA were used for retrieving fungi. At least three dilution series were used for each sample. Each vacuum dust sample was sieved through a 250 mm sieve. Approximately 100 mg of fine dust (< 250 mm) retrieved were used for fungal analysis by aforementioned dilution plating.

All plates were incubated in a 25°C incubator. They were examined every other day for up to 10 days to ensure the full recovery of fungi. Fungal identification was based on colony morphology, spores and conidia formation. Total fungal colonies formed on each MEA plate and *Stachybotrys chartarum* on CCA plates were counted and recorded. Fungal levels in samples were presented as colony forming units (CFUs) per measuring unit. For example, CFU/m^3 for Andersen® air samples, CFU/in^2 for wipe samples, CFU/plate for contact plate samples, and CFU/g of fine dust for vacuum dust samples.

All Zefon® cassette samples were analyzed by the Environmental Microbiology Laboratory in Escondido, California for direct microscopic examination. Fungal spores were identified and their airborne levels were presented as spores/m^3.

RESULTS AND DISCUSSION

Temperature and Relative Humidity

Indoor temperature and relative humidity measurements ranged from 74.9°F to 79.2°F, and 17.3% – 19.2%, respectively (Table 1). Outdoors temperature reading was lower, but with a higher relative humidity.

Microbiological Analyses Results

All laboratory analytical reports from FOH's EML are presented in Attachment A in a laboratory report #NOAA-00-31R. Results from microscopic examination of Zefonâ cassette samples are presented in Attachment B.

Air Samples

Andersen Results

No fungal growth was detected from indoor air samples. Mean outdoor airborne fungal level was 206 CFU/m^3 (Table 1). *Cladosporium* was the predominant fungal genus detected outdoors. Other fungi detected were *Penicillium, Epicoccum,* and *Paecilomyces. Stachybotrys chartarum* was not detected from these samples.

Zefon Results

No fungal spores were detected from samples collected from 6747 and 6855. *Cladosporium* (27 spores/m^3) was detected from the sample collected from 6745. Outdoor fungal spore levels were higher than those of indoors (Table 1). Fungal spores detected from outdoors were *Cladosporium* and Basidiospores. *Stachybotrys chartarum* was not detected from any sample collected.

Table 1. Temperature and relative humidity measurements and airborne fungal levels at different rooms of the 6th floor in SSMC-3 on February 17, 2000.

Rooms / Parameters	6745	6747	6855	Outdoors
Temperature (°F)	79.2	77.7	74.9	47.5
Relative Humidity (%)	17.3	18.0	19.2	20.9
Airborne Fungal Levels (CFU/m^3)	<12	<12	<12	200* 212
Total Fungal Spores (Spores/m^3)	27	<7	<7	294* 280

* Two samples were collected from outdoors.

Swab Samples

Most (7 out of 9) samples collected from surfaces of supply diffusers and return troughers in light fixtures were below the detection limits (BDL) (3 CFU/in^2 for supply diffuser and 2 CFU/in^2 for return trougher). The samples showing fungal growth were collected from return trougher surfaces in room 6855 and 6747, respectively, with 2 CFU/in^2 of *Cladosporium* and *Rhizopus* (samples #W08 and W24). *Stachybotrys chartarum* was not detected.

Contact Plate Samples

In general, fungal levels on these surfaces were low, although higher fungal levels were detected from the horizontal surfaces than vertical surfaces (Table 2). Fungal levels ranged from BDL of 1 CFU/plate to 6 CFU/plate. *Cladosporium* was the predominant fungal genus recovered. Other fungi recovered were *Alternaria, Chaetomium, Penicillium, Paecilomyces, Aspergillus fumigatus,* Basidiomycetes, and yeast.

Vacuum Dust Samples

Plenum Dust

Fungal levels in the fine dust collected from the plenum were at 10^3 - 10^4 CFU/g of fine dust levels (Table 3). *Penicillium* and *Aspergillus niger* were the predominant fungal genera detected from these samples, followed by *Cladosporium*. *Stachybotrys chartarum* was detected from room 6855 and the reference room 6745.

Furniture Dust

Fungal levels in the fine dust in furniture of these rooms were at the levels of 10^3 CFU/g of fine dust (Table 3). Predominant fungi detected were *Alternaria, Aureobasidium,* and *Cladosporium. Stachybotrys chartarum* was detected from furniture dust samples collected from these three rooms (Table 3).

Carpet Dust

Diverse fungal genera such as *Cladosporium, Aspergillus, Aureobasidium, Epicoccum, Paecilomyces, Penicillium,* and yeast were recovered from carpet dust samples. Fungal levels in the fine dust in carpeting of these rooms were at the levels of 10^3 – 10^4 CFU/g of fine dust (Table 3). *Stachybotrys chartarum* was detected from room 6747 and the reference room 6745 (Table 3).

Table 2. Fungal levels (CFU/plate) on horizontal and vertical surfaces of different rooms at the 6th floor of SSMC-3, by contact plate sampling collected on February 17, 2000.

Rooms **Parameters**	**6745**	**6747**	**6855**

Horizontal Surfaces (CFU/plate)	1 – 6* (3**)	1 – 4 (3)	<1 – 4 (3)
Vertical Surfaces (CFU/plate)	<1 (1)	1 (2)	<1 – 2 (2)

* Ranges. ** Total sample number.

Table 3. Total fungal levels (CFU/g of fine dust) in fine dust collected from carpet, plenum, and furniture of rooms 6745, 6747, and 6855 of SSMC-3, by vacuum dust sampling, collected on February 17, 2000.

Rooms / Parameters	**6745**	**6747**	**6855**
Plenum (CFU/g of fine dust)	4,752 (+*)	8,317 (-)	17,426 (+)
Carpet (CFU/g of fine dust)	5,200 (+)	13,861 (+)	5,545 (-)
Furniture (CFU/g of fine dust)	2,500 (+)	8,986 (+)	4,828 (+)

* +: *Stachybotrys chartarum* was detected on MEA and/or CCA plates.

-: *Stachybotrys chartarum* was not detected on MEA and CCA plates.

CONCLUSIONS

· No fungal growth was detected from indoor Andersen samples. Indoor fungal spore levels, by Zefon sampler, were lower than those of outdoors.

· In general, fungal burden on surfaces was low.

· *Stachybotrys chartarum* was not detected from any air, wipe, or contact plate samples collected.

· Very low fungal burden was detected from wipe samples collected from surfaces of supply diffusers and return troughers in light fixture.

· Fungal levels in plenum, carpet, and furniture dust of these rooms were at 10^3 - 10^4 CFU/g of fine dust levels. *Stachybotrys chartarum* was detected from all samples except for the plenum dust collected from room 6747 and carpet dust from room 6855.

· A diverse fungal population was recovered from these dust samples. *Penicillium* and *Aspergillus niger* dominated dust samples collected from the ceiling plenum.

RECOMMENDATIONS

· Conduct thorough HEPA vacuuming of furniture and carpeting in these rooms.

· Conduct any above ceiling plenum work after office hours. Thoroughly HEPA vacuum the surrounding areas afterwards.

· Implement an emergency water intrusion protocol for this building to adequately manage any unexpected water intrusion in order to prevent fungal proliferation.

ATTACHMENT A

USPHS DFOH ENVIRONMENTAL MICROBIOLOGY LABORATORY, PHILADELPHIA, PA

LABORATORY REPORT #NOAA-00-31R

Client agency: National Oceanic and Atmospheric Administration, Silver Spring, MD

POIS#/task #: D8H00CO31200 / 9903

Sampling date: 2/17/00

Dates of inoculation: 2/17/00 (airs and contact plates), 2/18/00 (wipes), and 2/19/00 (dust)

General location: SSMC-3, Silver Spring, MD

Specific location: 6th floor

Sampling techniques: Air (Andersen N-6 sampler), contact plate, wipe, and vacuum dust samplings

Medium used: Malt extract agar (MEA) and cellulose Czapek agar (CCA) for fungi

Samples submitted by: J. Sobelman

Date characterization completed: 2/29/00

(A) Air samples on MEA and CCA plates

Sample ID	Sampling Location	Air Volume (L)	Fungi on MEA @ 25º C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C

4-6855-0217A1, A2	6th floor, room 6855, cube	84.9	**No fungal growth** CFU/m^3 < 12	No
4-6745-0217A1, A2	6th floor, room 6745, center of cube	84.9	**No fungal growth** CFU/m^3 < 12	No
4-6747-0217A1, A2	6th floor, room 6747, center of cube	84.9	**No fungal growth** CFU/m^3 < 12	No
3-OA1-0217, 3-OA2-0217	Outside bldg. 3	84.9	1. *Cladosporium* (13*) 2. *Penicillium* (2) 3. *Epicoccum* (1) 4. *Paecilomyces* (1) CFU/m^3 = 200	No
3-OA1-0217, 3-OA2-0217	Outside bldg. 3	28.3	1. *Cladosporium* (5) 2. Basidiomycetes (1) CFU/m^3 = 212	No
FB	Field blank	NA#	No fungal growth	No

Sample ID	Sampling Location	Air Volume (L)	Fungi on MEA @ 25º C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
SB	Shipping blank	NA	No fungal growth	No

(B) Contact plate samples on MEA plates

Sample ID	Sampling Location	Fungi detected on MEA @ 25º C
4-6855-0217CP1	6th floor, room 6855, wall near window	**1. *Chaetomium* (1)** 2. *Cladosporium* (1) **CFU/plate = 2**
4-6855-0217CP2	6th floor, room 6855, top of desk	**No fungal growth** CFU/plate < 1

4-6855-0217CP3	6th floor, room 6855, top of system furniture	**1. *Cladosporium* (2)** 2. *Penicillium* (1) 3. Basidiomycetes (1) **CFU/plate = 4**
4-6855-0217CP4	6th floor, room 6855, top of grey file cabinet	**1. *Cladosporium* (2)** 2. *Penicillium* (1) 3. Basidiomycetes (1) **CFU/plate = 4**
4-6855-0217CP5	6th floor, room 6855, front of grey file cabinet	**No fungal growth** CFU/plate < 1
4-6745-0217CP1	6th floor, room 6745, wall column	**No fungal growth** CFU/plate < 1
4-6745-0217CP2	6th floor, room 6745, top of desk	**1. *Cladosporium* (2)** 2. *Penicillium* (1) **CFU/plate = 3**

Sample ID	Sampling Location	Fungi detected on MEA @ 25º C
4-6745-0217CP3	6th floor, room 6745, top of system furniture	**1. *Cladosporium* (3)** 2. *Aspergillus fumigatus*** (1) 3. *Penicillium* (1) 4. Basidiomycetes (1) **CFU/plate = 6**
4-6745-0217CP4	6th floor, room 6745, top of computer	1. *Alternaria* (1) **CFU/plate = 1**
4-6747-0217CP1	6th floor, room 6747, top of desk	1. *Penicillium* (1) **CFU/plate = 1**
4-6747-0217CP2	6th floor, room 6747, top of system furniture	1. *Penicillium* (1) 2. Basidiomycetes (1) **CFU/plate = 2**
4-6747-0217CP3	6th floor, room 6747, front of file	1. *Paecilomyces* (1) **CFU/plate = 1**

4-6747-0217CP4	6th floor, room 6747, top of scanner	1. *Cladosporium* (2) 2. yeast (2) **CFU/plate = 4**
4-6747-0217CP5	6th floor, room 6747, front of CPU	1. yeast (1) **CFU/plate = 1**

(C) Wipe samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
Blank	Blank	Blank	NA	10X-MEA 10X-CCA	No fungal growth	No

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
W08	4-6855-0217R1	6th floor, room 6855, return	5	10X-MEA 10X-CCA	1. *Cladosporium* (1) CFU/in^2 = 2	No
W09	4-6855-0217S1	6th floor, room 6855, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W10	4-6855-0217S2	6th floor, room 6855, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W19	4-6745-0217R1	6th floor, room 6745, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W20	4-6745-0217R2	6th floor, room 6745, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W21	4-6745-0217R3	6th floor, room 6745, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W22	4-6745-0217R4	6th floor, room 6745, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W23	4-6747-0217R1	6th floor, room 6747, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W24	4-6747-0217R1	6th floor, room 6747, return	5	10X-MEA 10X-CCA	1. *Rhizopus* (1) CFU/in^2 = 2	No

(D) Vacuum dust samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25° C
V01	4-6855-0217V01	6th floor, room 6855, furniture	0.029##	40X-MEA 10X-CCA	**1. *Aureobasidium* (2)** 2. *Alternaria* (1) 3. *Aspergillus niger*** (1) 4. *Epicoccum* (1) 5. *Rhizopus* (1) 6. *Stachybotrys chartarum**** (1) **CFU/g = 4,828**	Yes (9) CFU/g = 1,552
V02	4-6855-0217V02	6th floor, room 6855, carpet	0.101	40X-MEA 10X-CCA	**1. *Cladosporium* (6)** 2. *Aspergillus niger*** (2) 3. *Rhizopus* (2) 4. *Aspergillus fumigatus*** (1) 5. *Aspergillus sp.* (1) 6. *Paecilomyces* (1) 7. *Penicillium* (1) **CFU/g = 5,545**	No
V03	4-6855-0217V03	6th floor, room 6855, above ceiling	0.101	40X-MEA 10X-CCA	**1. *Penicillium* (32)** 2. *Aspergillus niger*** (8) 3. *Paecilomyces* (2) 4. *Chaetomium* (1) 5. *Epicoccum* (1) CFU/g = 1.7×10^4	Yes (1) CFU/g = 99
V04	4-6745-0217V01	6th floor, room 6745, furniture	0.056##	40X-MEA 40X-CCA	**1. *Alternaria* (4)** 2. *Aspergillus niger*** (2) 3. *Aureobasidium* (1) **CFU/g = 2,500**	Yes (1) CFU/g = 357

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
V05	4-6745-0217V02	6th floor, room 6745, carpet	0.100	40X-MEA 40X-CCA	**1. *Penicillium* (3)** 2. *Aureobasidium* (2) 3. *Epicoccum* (1) 4. yeast (7) **CFU/g = 5,200**	Yes (1) CFU/g = 400
V06	4-6745-0217V03	6th floor, room 6745, above ceiling	0.101	40X-MEA 10X-CCA	**1. *Aspergillus niger*** (7)** 2. *Cladosporium* (3) 3. *Penicillium* (2) **CFU/g = 4,752**	Yes (3) CFU/g = 297
V07	4-6747-0217V01	6th floor, room 6747, furniture	0.069##	40X-MEA 10X-CCA	**1. *Alternaria* (12)** 2. *Cladosporium* (7) 3. *Aspergillus niger*** (5) 4. *Bipolaris* (2) 5. *Epicoccum* (2) 6. *Penicillium* (2) 7. *Aureobasidium* (1) **CFU/g = 8,986**	Yes (1) CFU/g = 72
V08	4-6747-0217V02	6th floor, room 6747, carpet	0.101	40X-MEA 40X-CCA	**1. *Aureobasidium* (3)** 2. *Epicoccum* (3) 3. *Aspergillus niger*** (2) 4. yeast (27) CFU/g = 1.4×10^4	Yes (4) CFU/g = 1,584

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C

V09	4-6747-0217V03	6th floor, room 6747, above ceiling	0.101	40X-MEA 10X-CCA	**1. *Aspergillus niger*** (7)** 2. *Penicillium* (6) 3. *Cladosporium* (4) 4. *Alternaria* (3) 5. *Paecilomyces* (1) **CFU/g = 8,317**	No

* Colony counts.

** Opportunistic fungi.

*** Toxigenic fungi.

Not applicable.

5ml of sterilized distilled water were added instead of 10ml.

Microbiological laboratory report for samples collected

from the sixth floor of SSMC-3, on February 17, 2000.

ATTACHMENT B

Results from microscopic examination of Zefon air samples collected from the sixth floor of SSMC-3, on February 17, 2000.